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# PRELIMINARY EVALUATION OF SEED FUNGI FROM SEEDLOTS COLLECTED BY HAND PICKING AND BY NETTING IN 3 FEDERAL SEED ORCHARDS

by

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*Abstract*

*A variety of fungi were found common on the surface and under the seed coat of net-retrieved seed and seed from hand-picked cones. Few pathogenic fungi were recovered. Overall, seed from net retrieval had more fungi on the surface and under the seed coat than seed from hand-picked cones. Few fungi were found in seed endosperms, but more were found in seed from hand-picked cones than from net retrieval.*

## INTRODUCTION

Southern forestry has become increasingly dependent on a continuous supply of pine seed produced in intensively managed seed orchards. The cost of producing this seed is high because of the high cost of the various cultural practices necessary to obtain maximum production. Insects have been recognized as causing major seed losses but little is known about losses due to fungi. Seed fungi have long been recognized as a problem in agronomy. In 1977, Miller and Bramlett reported that pathogenic fungi were damaging slash pine seed. This led to several surveys of seed grown in southern seed orchards. Anderson and others (1981) found that in slash pine about 10 percent of sound seed and 15 percent of unsound seed contained potentially pathogenic internal fungi. Diplodia sp. was the most common pathogenic fungus isolated, followed by Fusarium sp. In one of the 2 years of the survey, Fusarium moniliforme var. subglutinans

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(causal agent of pitch canker) was recovered. In 1982, a survey of loblolly pine seed was made (Anderson, et.al. 1983) and pathogenic fungi were found on the surface of the seed in most seedlots. These fungi could be controlled with hydrogen peroxide. Only 2 of the 37 seedlots had enough internal fungi to be of any concern. Three of 12 slash pine seedlots from Texas yielded high levels of the potentially pathogenic fungi Botryodiplodia sp. and Fusarium sp. (Covington and others 1982). The fungi were recovered from under the seedcoat and from endosperm tissue.

More recently seed fungi on seedlots from cones on the trees and on the ground were compared (Anderson et.al. 1983). Pathogenic fungi were common on the seed surface in both treatments, however, none of the seedlots had enough internal pathogenic fungi to be of concern. Overall, seed from cones collected on the ground had more external and internal fungi.

The federal seed orchards in Region 8 are currently phasing in a new seed collection method which involves rolling out plastic netting on the ground, letting seeds fall naturally, and gathering them from the netting. The purpose of this evaluation was to make a preliminary determination of the differences in external and internal seed fungi isolated from net collected seed versus seed from hand picked cones.

## METHODS

Samples of net-collected seed were taken from the following sources:

| Orchard | Species | Source |
|---|---|---|
| Ouachita | shortleaf | East Ouachita |
| Stuart | loblolly | Louisiana loblolly |
| Erambert | loblolly | Alabama loblolly |

Seed from hand picked cones was obtained from a 15 clone mix (4 trees/clone) of the same sources acquired during insect and inventory monitoring activities. The clones represented in each source are listed below.

| | |
|---|---|
| East Ouachita | 2,8,11,20,22,23,28,32,34,38,39,43,46,47,49 |
| Louisiana loblolly | 1,5,6,11,15,16,19,22,25,30,34,35,37,42,47,49 |
| Alabama loblolly | 6,8,9,11,16,17,21,25,27,34,39,45,47,48,49,50 |

From each seedlot a sample of 125 seed was processed in three treatments; 1) 25 whole seeds were plated directly onto acidified (pH=4.7 via 50% lactic acid) potato dextrose agar (A-PDA); 2) 50 whole seeds were plated on A-PDA after being soaked 10 minutes in 20 percent hydrogen peroxide and rinsed with sterile, distilled water; and 3) 50 whole seed were soaked in 70 percent ethyl alcohol for 10 minutes, split, and one-half of the gametophyte tissue was removed and plated onto A-PDA. Seed in treatment 3 were separated as empty, sound and unsound. A sound seed appeared normal inside, while unsound gametophyte tissue appeared discolored or dried up.

Each plate was examined frequently for two weeks and isolated fungi identified.

## RESULTS

Results of culturing sample seeds are presented in tables 1 and 2. All seed had external fungi (treatment 1) while in treatment 2 the percent with fungi ranged from 18 - 100. Percent fungi in seed endosperms (treatment 3) ranged from 0-24.

Seed collected from the netting retrieval system had more fungi associated with the seed surface and the seed coat (treatments 1 and 2) than seed extracted from hand-picked cones. Increases were recorded in the number of fungal genera isolated (except Stuart treatment 2), in the total number of isolations recorded, and in the total percent of seed with fungi (in treatment 2). In treatment 3 fewer fungi were recovered from seed endosperms in all 3 orchards. Fewer fungal genera were represented (except Erambert), fewer total isolations and a lower percent total fungi.

Pathogenic fungi were recovered only in treatments 1 and 2. However, no real difference was apparent between seed from hand-picked cones or net retrieval. In one orchard more pathogens were present on net retrieval seeds while fewer were present in another. In other cases, isolations were equal or only slightly different. Almost all pathogenic fungi were Fusarium sp. or FMS. One isolation each of Phoma and Sphaeropsis was made. These might have potential to be pathogenic, however, this is at present only speculation.

## DISCUSSION

Since there appeared to be no definite increase in pathogenic fungi in treatments 1 and 2 and since the increase in fungi in treatment 2 on net-retrieved seeds was due primarily to non-pathogenic fungi, it appears that the net retrieval system has no serious detrimental effect on the populations of fungi associated with seed. Internal fungi were at a very low level regardless of collection method and in fact were less prevalent in net-collected seed.

Any seed treatment applied to rid seed of external fungi will also eliminate any difference between surface fungal populations of seed from hand-picked cones or net collected seed. It should be remembered, however, that this is a very limited survey of only 3 orchards and 1 seedlot in each. Variations in weather, site, species, pathogen populations, etc. could drastically affect the results of similar surveys. Conclusions should, therefore, be considered and preliminary despite similar results obtained from a survey of seed from hand-picked cones and ground-collected cones of loblolly pine (Anderson, et.al. 1983).

Table 1. Percent pathogens, other fungi and total fungi isolated from seed collected on netting and from hand-picked cones in 3 Federal Seed Orchards.

| Seedlot[a] | Treatment #1 | | | | Treatment #2 | | | | Treatment #3 | | | | All Treatment | | | | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|---|
| | % Known Pathogens | | % Other Fungi | Total % Fungi | % Known Pathogens | | % Other Fungi | Total % Fungi | % Known Pathogens | | % Other Fungi | Total % Fungi | % Known Pathogens | | | | % Other Fungi | Total % Fungi |
| | 1 | 2 | | | 1 | 2 | | | 1 | 2 | | | 1 | 2 | 3 | 4 | | |
| SM | 18 | 12 | 70 | 100 | 3 | 0 | 97 | 56 | 0 | 0 | 100 | 6 | 12 | 7 | 0 | 0 | 81 | 45 |
| NS | 25 | 10 | 65 | 100 | 3 | 0 | 97 | 94 | 0 | 0 | 0 | 0 | 14 | 5 | 0 | 0 | 81 | 58 |
| EM | 11 | 0 | 89 | 100 | 0 | 0 | 100 | 98 | 0 | 0 | 100 | 24 | 5 | 0 | 0 | 0 | 95 | 69 |
| NE | 8 | 1 | 91 | 100 | 0 | 1 | 98 | 100 | 0 | 0 | 100 | 6 | 4 | 1 | 0 | 0 | 95 | 62 |
| OM | 0 | 0 | 100 | 100 | 0 | 0 | 100 | 18 | 0 | 0 | 100 | 4 | 0 | 0 | 0 | 0 | 100 | 29 |
| NO | 0 | 0 | 100 | 100 | 1 | 0 | 99 | 100 | 0 | 0 | 0 | 0 | 1 | 0 | 0 | 0 | 99 | 60 |
| All picked | 11 | 4 | 85 | 100 | 1 | 0 | 99 | 57 | 0 | 0 | 100 | 11 | 7 | 2 | 0 | 0 | 91 | 47 |
| All netting | 10 | 3 | 87 | 100 | 1 | 0 | 99 | 98 | 0 | 0 | 100 | 2 | 6 | 2 | 0 | 0 | 92 | 67 |

KEY

1 - Fusarium sp.
2 - Fusarium moniliforme var. subglutinans

[a] SM = Stuart picked cones; NS = Stuart net-retrieved; EM = Erambert picked cones; NE = Erambert net-retrieved; OM = Ouachita picked cones; NO = Ouachita net-retrieved.

Table 2. Percent fungi number of genera and number of isolations of fungi from seed collected on netting and from hand-picked cones in 3 Federal Seed Orchards.

| | Treatment #1 | | | Treatment #2 | | | Treatment #3 | | | All Treatments | | |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Seedlot[a] | % Fungi[b] | No. Genera[c] | No. Isolates[d] | % Fungi | No. Genera | No. Isolates | % Fungi | No. Genera | No. Isolates | % Fungi | No. Genera | No. Isolates |
| SM | 100 | 8 | 51 | 56 | 9 | 29 | 6 | 3 | 3 | 45 | 14 | 83 |
| NS | 100 | 12 | 61 | 94 | 9 | 59 | 0 | 0 | 0 | 58 | 17 | 120 |
| EM | 100 | 6 | 56 | 98 | 5 | 54 | 24 | 2 | 12 | 69 | 7 | 122 |
| NE | 100 | 16 | 71 | 100 | 10 | 85 | 6 | 2 | 3 | 62 | 19 | 159 |
| OM | 100 | 2 | 29 | 18 | 1 | 9 | 4 | 1 | 2 | 29 | 3 | 40 |
| NO | 100 | 8 | 73 | 100 | 10 | 74 | 0 | 0 | 0 | 60 | 13 | 147 |
| All picked | 100 | 8 | 136 | 57 | 11 | 92 | 11 | 4 | 17 | 47 | 14 | 245 |
| All netting | 100 | 19 | 205 | 98 | 17 | 218 | 2 | 2 | 3 | 67 | 26 | 426 |

[a] SM = Stuart picked cones; NS = Stuart net-retrieved; EM = Erambert picked cones; NE = Erambert net-retrieved; OM = Ouahita picked cones; NO = Ouachita net-retrieved.
[b] Total percent of tested seeds yielding at least 1 fungus.
[c] Total number of fungal genera isolated.
[d] Total number of isolates of fungi recovered.

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